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## A STUDY OF SYSTEMATIC WOOD ANATOMY IN CANNABIS

BY  
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*Cannabis* has been associated with man since very early times (Ash, 1948), yet, surprisingly, little is known about its comparative wood anatomy. The reasons are due probably to the tendencies for (1) anatomists to select wood from trees and woody shrubs rather than from herbs for study and for (2) researchers often to disregard or slight plants associated with man, either as crops or weeds, in basic scientific enquiries.

Tippo (1938) offered a few general comments on the wood of *C. sativa* L. in his extensive study on the anatomy of the Moraceae and its allies. Stem shape and leaf-trace number in transections were stressed by Nasonov (1940) in a report on geographical races of hemp. Hayward (1948) devoted a chapter in his textbook to *C. sativa*. The general morphology of that species was given, but details of seedling anatomy and floral structure were emphasized; wood anatomy was scarcely mentioned. Metcalfe and Chalk (1950) summarized anatomical data on Cannabaceae to that date. Shimomura *et al.* (1967) emphasized trichomes in their study of leaf and bract anatomy in *Cannabis*; they found differences between *C. sativa* and *C. indica*.

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Some features of *Cannabis* anatomy are relatively well known, such as the economically important phloem (bast) fibres. These aspects have been reviewed by Hayward, 1948; Metcalfe and Chalk, 1950. Considerable attention has also been given to cystolithic hairs (Pireyre, 1961) and laticifers in *Cannabis*.

With the recent attention devoted to taxonomic problems in *Cannabis* (Schultes *et al.*, 1974; Stearn, 1974), I am pleased to present this introductory account on comparative wood anatomy. It includes apparently the first technical description of wood identified with vouchered material as *C. indica* Lam.

#### METHODS AND MATERIALS

All materials were collected fresh and preserved in formalin-propriono-alcohol (FPA). Woods were sectioned on a sliding microtome at 20  $\mu$ . Some sections of each sample were stained in safranin O and counterstained with fast green FCF and orange G; others were stained only with safranin. Tissues were mounted in Permout.

Xylem features were microscopically measured with a calibrated ocular micrometer; a minimum of 50 measurements were made for each feature reported in Table 1. Polarizing filters aided study of cell wall structure and crystals. Statistical analyses were made on a Wang 600 computer with the assistance of Dr. M. P. Johnson.

The material of *C. indica* came from a wild population at Pashimool, west of Kandahar, Afghanistan, *R. E. Schultes 26505* (Econ. Herb. Oakes Ames); that of *C. sativa* came from a naturalized population in Pottawatomie County, Kansas, United States, *L. C. Anderson 3663* (Fla. State Univ.).

#### RESULTS

Details of wood anatomy are illustrated in Figs. 1-6.

The woods of *C. indica* and *C. sativa* differ significantly in each feature listed in Table 1.

Vessels in *C. indica* tend to occur in radial chains; whereas those of *C. sativa* usually occur singly (as illustrated in Hayward, 1948). That difference in distribution can be seen in Figs. 1–2. Vessel members are angular to round in transection. They have simple perforation plates, and the end walls are slightly oblique. Pits are alternate with elliptic borders. Pit apertures are elongate; they are 6–9  $\mu$  long in *C. indica* and 4–8  $\mu$  in *C. sativa*.

Vessel members and wood fibres differ between the two samples in average width, length and cell wall thickness (Table 1). In *C. indica*, both cell types are wider, have thicker walls, but are shorter in length compared to those of *C. sativa*.

Fibres in the secondary xylem must not be confused with the hemp fibres of commerce, which are phloem or bast fibres. Wood fibres of *C. indica* are typical, lignified libriform fibres. Fibres in *C. sativa* differ in two respects. They are dimorphic, with successive tangential bands of

TABLE 1. Averaged measurements on wood anatomy in *Cannabis*.

Feature	<i>C. indica</i>	<i>C. sativa</i>	Significance level <sup>a</sup>
vessel number per group	3.05	1.39	*
vessel member width, $\mu$	68.52	62.16	*
vessel member wall thickness, $\mu$	3.50	2.30	**
vessel member length, $\mu$	209.71	244.54	**
fibre width, $\mu$	18.41	14.28	**
fibre wall thickness, $\mu$	3.44	0.68	*
fibre length, $\mu$	281.10	443.47	**
ray width (cell number)	2.23	1.63	**
ray height, mm	0.87	0.68	*

<sup>a</sup>Analysis of variance (F test): \* =  $p < .05$ , \*\* =  $p < .001$

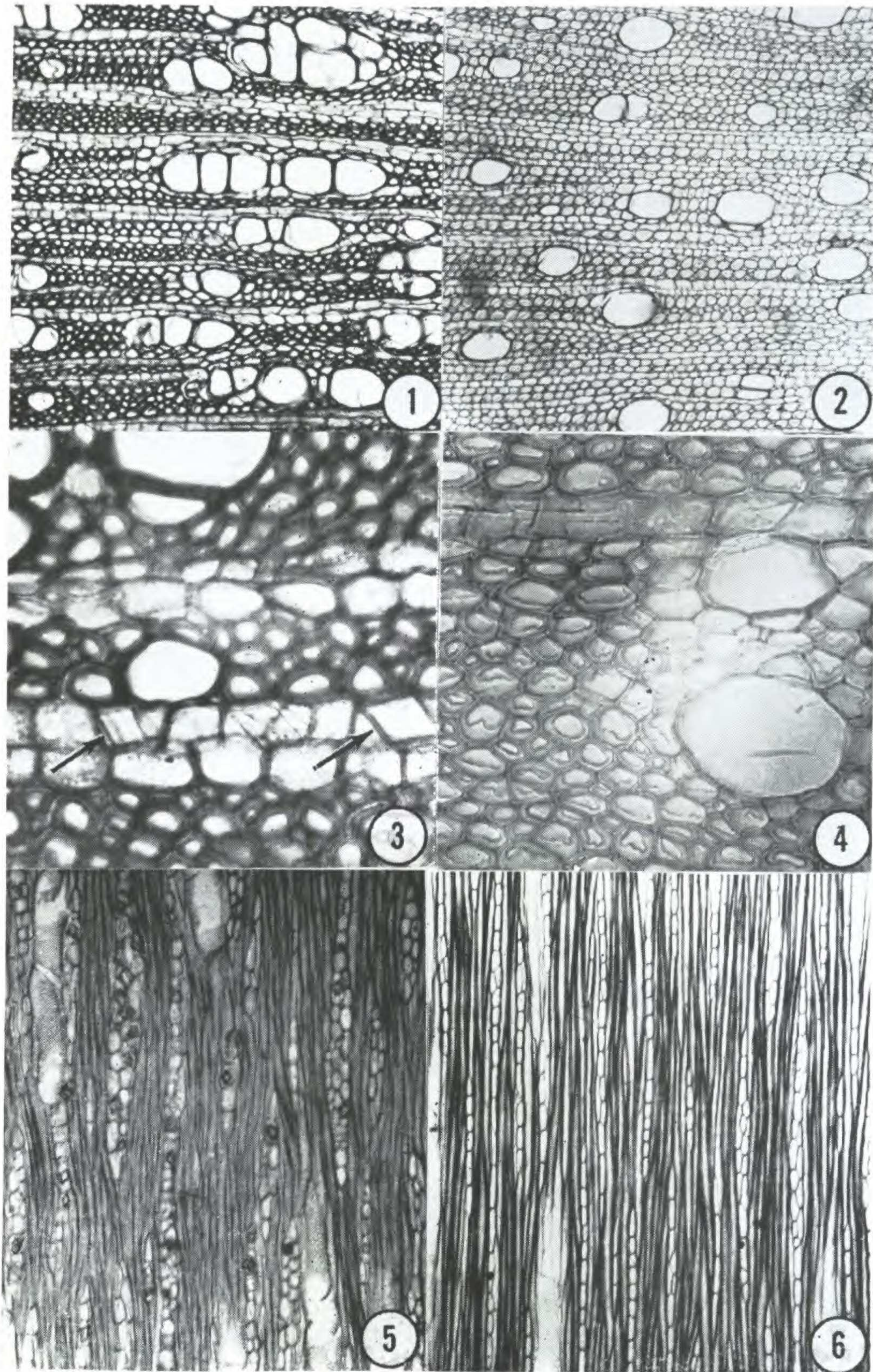
### EXPLANATION OF PLATE X

Figs. 1-6. *Cannabis* wood sections. Figs. 1, 3, 5 are *C. indica*, and 2, 4, 6 are *C. sativa*. Fig. 1, transection showing radial chains of vessels, libriform fibres, and procumbent ray parenchyma. Fig. 2, transection showing tendency for solitary vessels, fibre dimorphism, and ray parenchyma shorter radially (erect). Fig. 3, transection showing thick walls of vessel members and libriform fibres; note cuboidal crystals in ray cells (arrows). Fig. 4, transection showing relatively thin cell walls; note shrunken secondary walls of gelatinous fibres. Fig. 5, tangential section showing wood rays with numerous crystals; photographed with partially polarized light. Fig. 6, tangential section showing relatively narrower wood rays with erect cells; crystals absent. Figs. 1-2, 5-6,  $\times 71$ . Figs. 3-4,  $\times 308$ .

PLATE X

*C. indica*

*C. sativa*



thick-walled fibres alternating with bands of thin-walled fibres. They have irregularly shrunken secondary walls (more pronounced in the thick-walled fibres) and are termed gelatinous (Fig. 4). Their staining reaction (note lighter tones in Figs. 2, 4, 6) and absence of birefringence under polarized light are similar to that of gelatinous fibres in other species that I have studied (Anderson, 1963, 1972).

Axial parenchyma is paratracheal. It is very scanty in *C. indica* and scanty to vasicentric in *C. sativa*.

Wood rays are classed as heterogeneous I: *i.e.*, both multiseriates and uniseriates occur, and they are composed of procumbent and erect ray cells. Those of *C. indica* are predominantly square to procumbent; whereas ray cells in *C. sativa* are mostly erect with very few square or procumbent ones. The differences in cell shape are suggested in Figs. 5–6, but they are best viewed in radial sections. A qualitative difference in wood rays is the presence of numerous cuboidal or prismatic crystals of calcium oxalate in *C. indica*. They can be seen in all sections under normal light but are more obvious with partial polarization of light (Fig. 5). No crystals were found in *C. sativa* ray cells (although both species have druses in their phloem and ground tissues).

## DISCUSSION

Many American botanists have thought *Cannabis* to be monotypic, possibly because only hemp, *C. sativa*, has been cultivated in this country. Most taxonomists who have studied the genus closely, however, recognize three species: *C. indica* Lam., *C. ruderalis* Janisch., and *C. sativa* L. (see Schultes *et al.*, 1974, for a review of the taxonomic history of the genus).

Data from wood anatomy have not hitherto been utilized in the taxonomy of *Cannabis*. Such data might

help resolve the question of species recognition in the genus. Nasonov's study (1940) is of little use, as he mentioned no binomials. He primarily studied variation in crop plants (all *C. sativa?*), where he identified three basic types of stem structure. He did note that wild and cultivated forms of hemp could not be distinguished clearly on the basis of anatomy of stem and bast fibres.

Wood features of *C. indica* and *C. sativa* listed in Table 1 are those commonly measured in comparative studies. They are all significantly different between the species with four at the 5% level and five at the 0.1% level! Additional differences in the axial and radial parenchyma systems are noted in the text. Woods of the two species are qualitatively distinct for libriform fibres versus gelatinous fibres and for presence of crystals in wood rays. Many examples of the taxonomic significance of crystals in woods have been noted (Bailey, 1961; Chattaway, 1955-56).

Although only one sample of each species is discussed here, the magnitude of differences between the two is impressive in a system as conservative as wood. In his exhaustive review on many aspects of wood science, Jane (1963) stated the following regarding taxonomic wood anatomy:

Wood structure is probably more conservative than floral structure, and specific differences, as determined by floral characters, are often not reflected in the secondary xylem. Indeed, it may be said that in general the distinguishing features of wood are at generic, rather than specific, level.

Certainly, the plants used in this study are of the same genus, but it is my opinion that they represent different species.

Examination of woods from three additional collections of North American *C. sativa* shows they are also distinct from the *C. indica* wood sample. All vary from

*C. indica* in the features listed in Table 1, with the exception of vessel member width. All three samples have gelatinous fibres. Crystals are absent in wood rays in two; only a few were found in the rays of the third sample. Complete data on these samples will be presented in the future as part of an expanded study on the wood anatomy of *Cannabis*.

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